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EXAMINER	
ZHOU, SHUBO	

ART UNIT	PAPER NUMBER
1631	

DATE MAILED: 09/09/2004

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/083,682	WOLFFE ET AL.	
	Examiner	Art Unit	
	Shubo (Joe) Zhou	1631	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *01 June 2004*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-124* is/are pending in the application.

4a) Of the above claim(s) *27-65 and 72-124* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-26 and 66-71* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☒ The specification is objected to by the Examiner.

10)☒ The drawing(s) filed on *07 June 2002* is/are: a)☒ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☒ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO-1449 or PTO/SB/08) Paper No(s)/Mail Date *8/6/02*.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☒ Other: *See Continuation Sheet*.

Continuation of Attachment(s) 6). Other: Illustration of digestion of methylated chromatin with DpnI.

DETAILED ACTION

Election/Amendments

1. Applicants' election, with traverse, of Group IX (claim 66) in the response filed 6/1/04 is acknowledged. The traversal is on the ground that there would be no search burden to the Office if groups I through VIII, or groups IX and X, or groups XII through XV are examined together. This is not found fully persuasive because groups III through VIII are methods involving different steps and reagents than those of groups I and II, and produce different results. For example, the invention of group III involves treating chromatin with a nuclease, and it is drastically different from the invention of group II where a methylase is used because a methylase just modifies/marks the nucleic acid of the chromatin by adding methyl groups, whereas the nuclease does not modify/mark the nucleic acids of the chromatin but rather fragment it. Similarly, the methods of groups XII through XV have different procedures and produce different results. A thorough search for these groups would require distinct search strategies because of their divergent subject matter. However, upon further consideration, it is noted that the invention of group I is generic to the invention of group II, and the inventions of groups IX and X are not distinct from each other and not distinct from the invention of group I because the library of group IX (claim 66) is produced by the method of group I. Thus, the inventions of groups I, II, IX and X (i.e. claims 1-26, and 66-71) will be considered. Claims 27-65, and claims 72-124 are withdrawn from further consideration pursuant to 37 CFR 1.142(b), as being drawn to nonelected inventions, there being no allowable generic or linking claim. Applicant timely traversed the restriction (election) requirement in the response filed 6/1/04. The requirement is still deemed proper and is therefore made FINAL.

M.S. Brunsman 06 August 2004

Applicants' amendments to the specification are also acknowledged and entered.

Declaration

2. The declaration filed 6/7/02 is defective. A new declaration in compliance with 37 CFR 1.67(a) identifying this application by application number and filing date is required. See MPEP §§ 602.01 and 602.02.

The declaration is defective because it is not in compliance with 37 CFR 1.42 and 1.64. The declaration bears the name for the deceased co-inventor, Allen P. Wolffe, but it is not clear who made the signature under his name on the declaration. 37 CFR 1.42 states that the legal representative of the deceased inventor may make the oath or declaration, and 37 CFR 1.64 requires that if the legal representative makes the declaration, the declaration shall state that the person is a legal representative, and the citizenship, residence, and mailing address of the legal representative.

Information Disclosure Statement

3. The Information Disclosure Statement filed 8/6/02 has been entered and the references therein have been considered except references BK-1, DE 19853398C1, and WO 00/31294, which are not in English and are lined-through on form PTO-1449. A copy of the initialed form of PTO-1449 is attached herein.

4. The citations/listings of publications and/or patents in various sections of the specification such as those on page 18, etc. are not a proper Information Disclosure Statement. 37 CFR 1.98(b) requires a list of all patents, publications, or other information submitted for consideration by the Office, and MPEP § 609 A(1) states, "the list may not be incorporated into

the specification but must be submitted in a separate paper." Therefore, unless the references have been cited by the examiner on form PTO-892, they have not been considered.

Specification

5. The specification is objected to because of the following:

6. The title of the invention is not descriptive. The claims are drawn to methods for isolating a collection of polynucleotides corresponding to the accessible regions cellular chromatin and the libraries of polynucleotides isolated thereby, whereas the title is directed to database of regulatory sequences, method of making and using same. A new title is required that is clearly indicative of the invention to which the claims are directed.

7. Applicant is reminded of the proper content of an Abstract of the Disclosure. In chemical patent abstracts for compounds or compositions, the general nature of the compound or composition should be given as well as its use, *e.g.*, "The compounds are of the class of alkyl benzene sulfonyl ureas, useful as oral anti-diabetics." Exemplification of a species could be illustrative of members of the class. For processes, the type reaction, reagents and process conditions should be stated, generally illustrated by a single example unless variations are necessary. In the instant case, claims are drawn to methods and product, but the Abstract on page 152 of the specification does not state the process conditions, steps, etc. and does not give the general nature of the claimed product, i.e. polynucleotide libraries as well as their use(s). Complete revision of the content of the abstract is required on a separate sheet.

8. It is noted that trademarks are used in this application, such as GENBANK™ (registered by United States Department of Health and Human Services) on page 26, line 21. Trademarks should be capitalized wherever it appears and be accompanied by the generic terminology.

Although the use of trademarks is permissible in patent applications, the proprietary nature of the marks should be respected and every effort made to prevent their use in any manner that might adversely affect their validity as trademarks.

9. The disclosure is objected to also because it contains an embedded hyperlink and/or other form or browser-executable code. Such code is present in the specification at page 26, line 21, and elsewhere. Applicants are required to delete the embedded hyperlink and/or other form of browser-executable code. See MPEP ' 608.01.

10. Appropriate correction is required.

Claim Rejections-35 USC § 112

11. The following is a quotation of the **first** paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

12. Claims 66-71 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. The claims contain subject matter that was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

The claims, as currently written, are drawn to library or libraries of polynucleotides comprising polynucleotides corresponding to the accessible regions of cellular chromatin obtained by the method of claim 3. Each of these claims is directed to a genus comprising any library of polynucleotides comprising polynucleotides corresponding to the accessible regions of cellular chromatin obtained by the method of claim 3. Note that absent an explicit definition in the specification of the term, a library is interpreted as "an unordered collection of clones (i.e., cloned DNA from a particular organism)" (see Biotech Life Science Dictionary, URL: http://biotech.icmb.utexas.edu/search/dict-search.phtml?title=library). Thus the number of clones of polynucleotides in each library may vary. Further, since the probes used in claim 3 may be a chemical, an enzyme or an antibody, each of which may react with, and thus mark, different polynucleotides, the claimed genus comprises different species of libraries comprising different polynucleotides.

A description of a genus may be achieved by means of a recitation of a representative number of species, falling within the scope of the genus, or by means of a recitation of structural features common to the members of the genus, which features constitute a substantial portion of the genus. Regents of the University of California v. Eli Lilly & Co., 119 F3d 1559, 1569, 43 USPQ2d 1398, 1406 (Fed. Cir. 1997). In the instant case, however, the specification does not describe the structure (i.e. the sequences of each clone of a library) of any species, nor does it describe any structural feature (i.e. the sequence of each clone in a library) common to the members of the genus. No common structural attributes identify the members of the genus. While the specification gives example of how to make a library (see pages 113-116), it does not describe the structure of the library or libraries made. The general knowledge and level of skill in

the art do not supplement the omitted description because specific, not general, guidance is what is needed. Since the disclosure fails to describe the common structural attributes or characteristics that identify members of the genus, and because the genus is highly variable, the mere example of a method to make a library is insufficient to describe the genus. One of skill in the art would reasonably conclude that applicant was not in possession of the claimed genus at the time the application was filed.

13. Claims 16-20 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the enablement requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention.

In *In re Wands* (8 USPQ2d 1400 (CAFC 1988)), the CAFC considered the issue of enablement in molecular biology. The CAFC summarized eight factors to be considered in a determination of "undue experimentation". These factors include: (a) the quantity of experimentation; (b) the amount of guidance presented; (c) the presence or absence of working examples; (d) the nature of the invention; (e) the state of the prior art; (f) the predictability of the prior art; (g) the breadth of the claims; and (h) the relative skill in the art. The factors are analyzed for the instant case as follows:

(a) In the instant case, the amount of experimentation required by the skilled artisan in order to practice of making and using the claimed methods would require an unpredictable amount of experimentation for the following reasons:

(b)-(c) The claims are drawn to a method of isolating a collection of polynucleotides

corresponding to accessible regions of cellular chromatin, the method comprising treating the cellular chromatin with a methylase, digesting the chromatin with a methylation-dependent restriction enzyme to produce a collection of fragments of methylated polynucleotides and non-methylated polynucleotides, and isolating a collection of the non-methylated polynucleotides, whereby the termini of the non-methylated polynucleotides correspond to accessible region of cellular chromatin. While the instant specification provides example of a method using a methylation-sensitive restriction enzyme to digest the methylated chromatin before isolating the methylated polynucleotide fragments, it does not present an example of a method where a methylation-dependent restriction enzyme is used to digest the methylated chromatin before isolating the un-methylated polynucleotide fragments. It also fails to provide a guidance that teaches the skilled artisan how to practice the claimed method.

(d)-(h) The nature of the invention, i.e. a method of isolating a collection of polynucleotides corresponding to accessible regions of cellular chromatin involving methylating chromatin, digesting the methylated chromatin with a methylation-dependent restriction enzyme and isolating the unmethylated fragments, is complex, and the prior art does not teach such a method. However, application of using a methylase to mark a region of chromatin, a methylation-dependent restriction enzyme to digest the methylated chromatin and isolation of the methylated fragments (not the unmethylated fragments as required in the instant claims) is taught or suggested by van Steensel et al. (Nature Biotechnology, Vol. 18, April 2000, pages 424-428). It is well known in the art that methylation-dependent restriction enzyme, e.g. Dpn I, recognizes a site of a double stranded DNA and cuts it only if the recognition site is methylated. See the general notes for Dpn I of the New England Biolabs Website (page 2 of 3 of the internet website

printout at <http://www.neb.com/nebecomm/products/productR0176.asp>). This is also admitted by applicants in the specification (see page 31, lines 30-32). Take Dpn I as an illustrating example (see the attached illustration for digestion of a methylated chromatin with Dpn I). Given that Dpn I only cuts the DNA if and when the "A"s on both strands of the recognition site 5'GATC3' are methylated, and that each cut results in two ends which both carry a methyl group and are thus both methylated, the digestion of a chromatin methylated at multiple portions of the accessible regions with DpnI would result in fragments which all carry at least one methyl group and are thus all methylated. Based on this analysis and the teachings of the prior art, it would be unconceivable to isolate any unmethylated fragments after such a digestion and for such fragments to correspond to the accessible regions of the chromatin.

The skilled practitioner would first turn to the instant specification for guidance in practice of the claimed method for isolating a collection of polynucleotides corresponding to accessible regions of cellular chromatin. However, the specification does not provide sufficient guidance of practicing the method. As such, the skilled practitioner would turn to the prior art for such guidance. However, the prior art does not teach the method, and the knowledge in the art would suggest the implausibility of the method. Finally, said practitioner would have to turn to trial and error experimentation for practicing the method without guidance from the specification or the prior art. Therefore, undue experimentation becomes the burden of the practitioner.

14. Claims 1-6, 8-15, and 21-26 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for practicing the claimed method involving using a probe

to mark the chromatin with any marks but methylation, or when marking the chromatin with methylation, digesting the methylated chromatin with a methylation-sensitive restriction enzyme and isolating the methylation-marked fragments, does not reasonably provide enablement for practicing the method involving methylating the chromatin, digesting the methylated chromatin with a methylation-dependent restriction enzyme. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to practice the invention commensurate in scope with these claims.

The claims are drawn to a genus where the chromatin are marked by different probes and the chromatin are fragmented with different means and either the marked or unmarked fragments are isolated to obtain polynucleotides corresponding to the accessible regions of the cellular chromatin. The claimed method encompasses an embodiment where cellular chromatin is methylated, which is then digested with methylation-dependent restriction enzyme, followed by isolation of the methylated polynucleotides to isolate the polynucleotides that correspond to the accessible regions of cellular chromatin. As shown in the attached illustration for digestion of a methylated chromatin with Dpn I, all the fragments resulted from the digestion are methylated. Thus, the methylated polynucleotides isolated by the claimed methods comprise the fragments of the whole chromatin not just those corresponding to the accessible regions, but also polynucleotides corresponding to the inaccessible regions of chromatin. For the status of the prior art, etc., see the analysis of the Wands factors as set forth above in the previous seciton.

15. The following is a quotation of the **second** paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

16. Claims 66-71 are rejected under 35 U.S.C. 112 , second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim 66 recites the limitation "wherein the polynucleotides are obtained according to claim 3" in lines 2-3; and claim 67 recites the limitation "the nucleotides in each library are obtained according to the method of claim 3" in lines 4-5. The metes and bounds of these limitations are not clear. There are different polynucleotides obtained in claim 3, marked polynucleotides, unmarked polynucleotides and the mixture thereof. It is unclear what polynucleotides among these from claim 3 are meant in the limitations of claims 66 and 67. Therefore the metes and bounds of the claimed invention are unclear.

Claims 68-71 are rejected because they depend from claim 67 and also contain the aforementioned indefinite limitation.

Claim Rejections-35 USC § 102

17. The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

18. Claims 66-71 are rejected under 35 U.S.C. 102(b) as being anticipated by Clontech (Clontech Catalog, 1998-1999, pages 177-183, Clontech Laboratories, Inc., Palo Alto,

California).

The claims are drawn to library or libraries comprising polynucleotides that correspond to accessible regions of cellular chromatin. Due to the "comprising" language in the claims, it is interpreted that the claimed library or libraries also comprise polynucleotides that correspond to the inaccessible regions of the cellular chromatin. Thus, the claims read on any complete genomic libraries of any organism with cellular chromatin because such libraries would comprise polynucleotides that do, and polynucleotides that do not correspond to the accessible regions.

The claims, as currently written, are apparently product-by-process claims.

The court in *In re Thorpe 777 F.2d 695, 698, 227 USPQ 964,966 (Fed. Cir. 1985)* holds:

> "[E]ven though product-by-process claims are limited by and defined by the process, determination of patentability is based on the product itself. The patentability of a product does not depend on its method of production. If the product in the product-by-process claim is the same as or obvious from a product of the prior art, the claim is unpatentable even though the prior product was made by a different process."

Clontech Catalog discloses multiple genomic libraries made from different organisms with cellular chromatin using different vector systems. See pages 177-183, especially the table on pages 182-183. These genomic libraries are made by a method involving digesting the whole genomes of the chromatin of different organisms with Sau3A I or Mbo I, which are four cutters and are known to digest the genomes frequently, and cloning the resulted fragments in different vector systems. See page 177. Therefore, it would be readily apparent to one of ordinary skill in the art that the libraries inherently comprise polynucleotides that correspond to accessible regions of cellular chromatin. Further, the catalog discloses a plurality of libraries comprising polynucleotides from cellular chromatin of cells at different stages of the development, such as

from mouse of ages of 9-11 weeks, and adult, and a plurality of libraries comprising polynucleotides from different tissues, such as mouse kidney and mouse liver. See the listing of genomic libraries on pages 182-183.The catalog also discloses a plurality of libraries comprising polynucleotides from healthy and diseased cells, such as normal muscle of Xenopus laevis and human Hela S3 cells (from ATCC#CCL2.2, see page 182), which are diseased (cancer) cells and infected with viruses. See page 1 of 3 of the printout of ATCC catalog from ATCC's website: http://www.atcc.org/SearchCatalogs/longview.cfm?atccsearch=yes.

Claim Rejections-35 USC § 103

19. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

20. The factual inquiries set forth in *Graham* **v.** *John Deere Co.*, 383 U.S. 1, 148 USPQ 459 (1966), that are applied for establishing a background for determining obviousness under 35 U.S.C. 103(a) are summarized as follows:

1. Determining the scope and contents of the prior art.
2. Ascertaining the differences between the prior art and the claims at issue.
3. Resolving the level of ordinary skill in the pertinent art.
4. Considering objective evidence present in the application indicating obviousness or nonobviousness.

21. This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various

claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

22. Claims 1-2, 4-9, 13-19, 21-25 are rejected under 35 U.S.C. 103(a) as being obvious over van Steensel et al. (Nature Biotechnology, Vol. 18, pages 424-428, April 2000, journal received by the STIC of the USPTO on 4/19/2000) in view of Bringmann et al. (FEBS Letters, Vol. 213, number 2, pages 309-315, 1987).

The claims are drawn to method of isolating a collection of polynucleotides corresponding to the accessible regions of cellular chromatin involving treating cellular chromatin with a probe that modifies or marks the accessible region of chromatin, fragmenting the treated chromatin to produce marked and unmarked polynucleotides, and isolating the marked polynucleotide fragments which correspond to the accessible region. It is pointed out that the treatment of chromatin with a probe in the claims, as written, can happen in vitro or in vivo.

Van Steensel et al. disclose a method of identifying regulatory DNA sequences that bind to proteins. Such sequences to which protein binds to are interpreted being sequences corresponding to the accessible regions of cellular chromatin because only if these sequences of chromatin are accessible to the protein, can it bind to the sequences. The method comprises introducing a protein comprising E. coli dam methylase into fruit fly Kc cells where the dam methylase modifies chromatin by methylating the N^6-position of adenine in the sequence GATC

(see page 424, Abstract, and left column), isolating genomic DNA and fragmenting the DNA with restriction enzymes DpnI or DpnII (DpnI is methylation-dependent and DpnII is methylation-sensitive. See printouts from New England Biolabs' website, page 2 of 3 for DpnI and page 2 of 3 for DpnII). The digested DNA fragments were used for identification of the sequences that are methylated by Southern blot analysis or for quantification of methylation by PCR. See page 427, right column, and page 428, left column. Note that introducing a dam methylase into the cell where the methylase reacts with chromatin is interpreted as being treating the cellular chromatin, as required in the claims.

Van Steensel et al. do not actually isolated the methylated (marked) polynucleotides from the unmethylated (unmarked) polynucleotides. However, they explicitly suggest doing so by stating that the methylated DNA fragments could be purified using an ^{m6}A-specific antibody and these fragments could be used as probes to screen DNA microarrays representing the whole genome to obtain regulatory DNA sequences genome-wide. See page 427, the paragraph bridging the left and right columns.

Bringmann et al. disclose a method of using an ^{m6}A-specific antibody to isolate ^{m6}A-methylated polynucleotides. See page 310.

One of ordinary skill in the art at the time the invention was made would have been motivated by van Steensel et al. to modified the method disclosed by the authors to further isolate the methylated, i.e. marked, DNA fragments by using the antibody suggested in order to perform a genome wide analysis. There would have been a reasonable expectation of success because Bringmann et al. provide the detailed procedures of the isolation process. Therefore, the

claimed invention would have been obvious to one of ordinary skill in the art at the time the invention was made.

In addition, as to claim 2, the chromatin is deproteinized with proteinase K. See page 428, left column.

In regard to claims 4, 7, 18, and 23, the fragmentation of chromatin/DNA is by restriction enzymes DpnI or DpnII which is methylation-sensitive. See page 427, right column and page 428, left column.

In regard to claims 5-6,13,17, and 22, the probe is an E. coli dam methylase, an enzyme, and the marked polynucleotides are methylated. See page 424, left column, bottom paragraph. Note that although the dam methylase used in the method is linked to another protein, the dam methylase functions normally.

In regard to claims 8, 19, and 24, the digested DNA was separated and fractionated by size on a 1.5% agarose gel and blotted to nylon membrane, where the DpnI released fragments, which are marked with methylase, were identified and quantified. See the bridging paragraph on pages 427-428.

In regard to claim 14, which requires that the probe is a chemical, due to the absence of an explicit definition for the term "chemical" in the specification, it is interpreted based on a dictionary definition as "A substance composed of chemical elements or obtained by chemical processes" (The On-line Medical Dictionary, URL: < http://cancerweb.ncl.ac.uk/cgi-bin/omd?chemical>. Since the methylase used in the method of van Steensel et al. is a protein composed of chemical elements or obtained by biochemical process, it is broadly interpreted that it is a chemical.

In regard to claims 15-20, which require also the isolation of the unmarked, i.e. the unmethylated polynucleotides fragments, it is pointed out that since the polynucleotide fragments after digestion by the restriction enzyme comprise the methylated and un-methylated polynucleotides, after the methylated fragments are isolated, the rest, i.e. the unmethylated fragments are also isolated (away from the methylated fragments).

It should also be pointed out that as to step (c) of claim 1, due to the absence of an explicit definition in the specification for the term "isolated" or "isolating" to define exactly away from what the polynucleotides are isolated, it could be broadly interpreted that the polynucleotides are isolated away from proteins, etc.

23. Claim 3 is rejected under 35 U.S.C. 103(a) as being obvious over van Steensel et al. (Nature Biotechnology, Vol. 18, pages 424-428, April 2000, journal received by the STIC of the USPTO on 4/19/2000) in view of Bringmann et al. (FEBS Letters, Vol. 213, number 2, pages 309-315, 1987), as applied to claims 1-2, 4-9, 13-19, 21-25 above, further in view of Grosveld et al. (US patent 5,635,355, Jun 3, 1997).

Claim 3 is drawn to a method of isolating a collection of polynucleotides corresponding to the accessible regions of cellular chromatin involving treating cellular chromatin with a probe that modifies or marks the accessible region of chromatin, fragmenting the treated chromatin to produce marked and unmarked polynucleotides, and isolating and cloning the marked polynucleotide fragments which correspond to the accessible region. It is pointed out that the treatment of chromatin with a probe in the claims, as written, can occur in vitro or in vivo (inside a cell or an organism).

As applied to claims 1-2, 4-9, 13-19, 21-25 above, Van Steensel et al. and Bringmann et al. teach a method of identifying regulatory DNA sequences that bind to proteins. The method comprises introducing a protein comprising E. coli dam methylase into fruit fly Kc cells where the dam methylase modifies chromatin by methylating the N^6-position of adenine in the sequence GATC (see page 424, Abstract, and left column), isolating genomic DNA and fragmenting the DNA with restriction enzymes DpnI or DpnII (DpnI is methylation-dependent and DpnII is methylation-sensitive. See printouts from New England Biolabs' website, page 2 of 3 for DpnI and page 2 of 3 for DpnII). The digested DNA fragments were used for identification of the sequences that are methylated by Southern blot analysis or for quantification of methylation by PCR. See page 427, right column, and page 428, left column. Van Steensel et al. also suggest isolating the methylated (marked) polynucleotides by using an ^{m6}A-specific antibody would bind to the methylated polynucleotides. However, Van Steensel et al. and Bringmann et al. do not explicitly teach or suggest cloning the isolated fragments.

Grosveld et al. teach a method of preparing nucleic acids which comprise regulatory sequences from a cell (see column 21, claim 1, line 1, for example). The method involves treating chromatin with a nuclease, fragmenting the chromatin with a restriction enzyme, and contacting DNA fragments of interests from precloned plasmids that contain Dnase I hypersensitive sites with a population of vectors to permit ligation of the DNA fragments in order to use it for in vivo introduction. See column 8, lines 1-25, and column 15, lines 43-47. So, Grosveld et al. expressly teach and suggest cloning of DNA fragments containing the Dnase I hypersensitive site, i.e. the regulatory sequences for in vivo usage.

One of ordinary skill in the art at the time of the invention was made would have been motivated by Grosveld et al. to modify the methods of van Steensel et al. and Bringmann et al. to further clone the isolated methylated polynucleotides for in vivo usage. There would have been a reasonable expectation of success because Grosveld et al. describe the vectors and other cloning reagents, and kits for cloning would also have been available and routine in the art. Therefore, there would have been a reasonable expectation of success.

24. Claims 12, 20, and 26 are rejected under 35 U.S.C. 103(a) as being obvious over van Steensel et al. (Nature Biotechnology, Vol. 18, pages 424-428, April 2000, journal received by the STIC of the USPTO on 4/19/2000) in view of Bringmann et al. (FEBS Letters, Vol. 213, number 2, pages 309-315, 1987), as applied to claims 1-2, 4-9, 13-19, 21-25 above, further in view of Gross (Ann. Rev. Biochem., 1988, 57:159-197).

The claims are drawn to a method of isolating a collection of polynucleotides corresponding to the accessible regions of cellular chromatin involving treating cellular chromatin with a probe that modifies or marks the accessible region of chromatin, fragmenting the treated chromatin to produce marked and unmarked polynucleotides, isolating and sequencing the marked polynucleotide fragments which correspond to the accessible region. It is pointed out that the treatment of chromatin with a probe in the claims, as written, can occur in vitro or in vivo (inside a cell or an organism).

As applied to claims 1-2, 4-9, 13-19, 21-25 above, Van Steensel et al. and Bringmann et al. teach a method of identifying regulatory DNA sequences that bind to proteins. The method comprises introducing a protein comprising E. coli dam methylase into fruit fly Kc cells where

the dam methylase modifies chromatin by methylating the N^6-position of adenine in the sequence GATC (see page 424, Abstract, and left column), isolating genomic DNA and fragmenting the DNA with restriction enzymes DpnI or DpnII (DpnI is methylation-dependent and DpnII is methylation-sensitive. See printouts from New England Biolabs' website, page 2 of 3 for DpnI and page 2 of 3 for DpnII). The digested DNA fragments were used for identification of the sequences that are methylated by Southern blot analysis or for quantification of methylation by PCR. See page 427, right column, and page 428, left column. Van Steensel et al. also suggest isolating the methylated (marked) polynucleotides by using an ^{m6}A-specific antibody would bind to the methylated polynucleotides. However, Van Steensel et al. and Bringmann et al. do not explicitly teach or suggest sequencing the isolated fragments.

Gross teaches in great length of the different aspects of nuclease hypersensitive sites which correspond to accessible regions of chromatin as well as their biological importance. See pages 159-187. Gross especially discusses the functional sequences associated with these regions such as enhancers, silencers, and promoters. See page 169 and page 170, Table 2. Gross expressly encourage obtaining the sequences of these nuclease hypersensitive sites including cis-acting sequences. He states that "knowledge of the cis-acting sequences and trans-acting factors that generate hypersensitive sites should become available in the future for a wide variety of important sequences"; "this information may permit the custom design of novel functional elements that override the problems of chromosomal position effects and misregulation often observed following the integration of transgenes", and "new approaches for gene therapy in medicine may even evolve from some of these principles." See page 188. This clearly stresses the importance of the knowledge of the exact sequences of the cis-acting elements, etc.

One of ordinary skill in the art at the time the invention was made would have been motivated by Gross to modify the methods of van Steensel et al. and Bringmann et al. to further sequence the isolated methylated polynucleotides in order to realize what Gross envisions -- permitting the custom design of novel functional elements that override the problems of chromosomal position effects and misregulation often observed following the integration of transgenes in gene therapy, etc. There would have been a reasonable expectation of success because the method of sequencing isolated DNA would have been available and routine in the art, e.g. Kinzler et al. (US patent 5695937, Dec 9, 1997) which provides detailed sequencing procedures. See also Tanguay et al. ((Nucleic Acids Research, Vol. 18, page 5902, 1990), which describe detailed procedures for cloning and sequencing fragments from enzyme digestions, page 5902.

25. Claims 1-4, 10-11, and 13-15 are rejected under 35 U.S.C. 103(a) as being obvious over Grosveld et al. (US patent 5,635,355, Jun 3, 1997) in view of Tanguay et al. (Nucleic Acids Research, Vol. 18, page 5902, 1990).

The claims are drawn to a method of isolating a collection of polynucleotides corresponding to the accessible regions of cellular chromatin involving treating cellular chromatin with a probe that reacted with the accessible region of chromatin, fragmenting the treated chromatin to produce marked and unmarked polynucleotides, isolating and/or cloning the marked polynucleotide fragments which correspond to the accessible region. It is pointed out that the treatment of chromatin with a probe in the claims, as written, can occur in vitro or in vivo (inside a cell or an organism).

Grosveld et al. teach a method of preparing nucleic acids which comprise regulatory sequences from a cell (see column 21, claim 1, line 1, for example). The method involves treating isolated nuclei which comprises chromatin with DNaseI, where the enzyme reacts with accessible regions of cellular chromatin (see column 8, lines 17-21), fragmenting the chromatin with a restriction enzyme to generate DNA fragments (column 8, lines 23-25, and contacting DNA fragments of interests from precloned plasmids that contain DNase I hypersensitive sites with a population of vectors to permit ligation of the DNA fragments in order to use it for in vivo introduction. See column 8, lines 1-25, and column 15, lines 43-47. Grosveld et al., however, do not clone the Dnase I hypersensitive fragments identified in column 8. However, Grosveld et al. expressly suggest cloning such fragments because they expressly claim "a method of obtaining a DNA fragment comprising a dominant activator sequence, comprising providing a candidate DNA fragment comprising a Dnase I hypersensitive site from a genetic locus" ..., and "ligating the fragment to an expressible gene to from a construct." Such Dnase I hypersensitive fragments should encompass fragments from readily available clones or from the fragments identified in column 8. See claim 1.

One of ordinary skill in the art would have been motivated to modify the method of Grosveld et al. to not only clone the Dnase I hypersensitive fragment from a readily made clone containing such fragment if the clone is available, but also to clone the Dnase I hypersensitive fragments directly from the fragments identified in column 8 if there are no clones readily available comprising such fragments in order to obtain such fragments which contain regulatory sequences. There would have been a reasonable expectation of success because such experimental methods of cloning and sequencing fragments of Dnase I digests would have been

routine. For example, Tanguay et al. teach of a method to directly clone and sequence the fragments of Dnase I digests. See page 5902.

As to claim 10, since the specification lacks an explicit definition for "isolated chromatin", it is interpreted that the chromatin in the isolated nuclei by Grosveld et al. is isolated chromatin because it is isolated away from the cellular membrane and away from the cytoplasm, etc.

26. Claim 12 is rejected under 35 U.S.C. 103(a) as being obvious over Grosveld et al. (US patent 5,635,355, Jun 3, 1997) in view of Tanguay et al. (Nucleic Acids Research, Vol. 18, page 5902, 1990), as applied to claims 1-4, 10-11, and 13-15 above, further in view of Gross (Ann. Rev. Biochem., 1988, 57:159-197).

The claim is drawn to a method of isolating a collection of polynucleotides corresponding to the accessible regions of cellular chromatin involving treating cellular chromatin with a probe that reacted with the accessible region of chromatin, fragmenting the treated chromatin to produce marked and unmarked polynucleotides, isolating and/or cloning and sequencing the marked polynucleotide fragments which correspond to the accessible region. It is pointed out that the treatment of chromatin with a probe in the claims, as written, can occur in vitro or in vivo (inside a cell or an organism).

As applied to claims 1-4, 10-11, and 13-15 above, Grosveld et al. teach a method of preparing nucleic acids which comprise regulatory sequences from a cell (see column 21, claim 1, line 1, for example). The method involves treating isolated nuclei which comprises chromatin with DNaseI, where the enzyme reacts with accessible regions of cellular chromatin (see column

8, lines 17-21), fragmenting the chromatin with a restriction enzyme to generate DNA fragments (column 8, lines 23-25, and contacting DNA fragments of interests from precloned plasmids that contain DNase I hypersensitive sites with a population of vectors to permit ligation of the DNA fragments in order to use it for in vivo introduction. See column 8, lines 1-25, and column 15, lines 43-47. Grosveld et al., however, do not clone the Dnase I hypersensitive fragments identified in column 8. However, Grosveld et al. expressly suggest cloning such fragments because they expressly claim "a method of obtaining a DNA fragment comprising a dominant activator sequence, comprising providing a candidate DNA fragment comprising a Dnase I hypersensitive site from a genetic locus" ..., and "ligating the fragment to an expressible gene to from a construct." Such Dnase I hypersensitive fragments should encompass fragments from readily available clones or from the fragments identified in column 8. See claim 1. Grosveld et al. do not explicitly teach of sequencing the fragments.

Gross teaches in great length of the different aspects of nuclease hypersensitive sites which correspond to accessible regions of chromatin as well as their biological importance. See pages 159-187. Gross especially discusses the functional sequences associated with these regions such as enhancers, silencers, and promoters. See page 169 and page 170, Table 2. Gross expressly encourage obtaining the sequences of these nuclease hypersensitive sites including cis-acting sequences. He states that "knowledge of the cis-acting sequences and trans-acting factors that generate hypersensitive sites should become available in the future for a wide variety of important sequences"; "this information may permit the custom design of novel functional elements that override the problems of chromosomal position effects and misregulation often observed following the integration of transgenes", and "new approaches for gene therapy in

medicine may even evolve from some of these principles." See page 188. This clearly stresses the importance of the knowledge of the exact sequences of the cis-acting elements, etc.

One of ordinary skill in the art at the time the invention was made would have been motivated by Gross to modify the methods of Grosveld et al. to further sequence the cloned polynucleotides in order to obtain the sequences and realize what Gross envisions -- permitting the custom design of novel functional elements that override the problems of chromosomal position effects and misregulation often observed following the integration of transgenes in gene therapy, etc. There would have been a reasonable expectation of success because the method of Tanguay et al. teaches of detailed procedures to directly clone and sequence the fragments of Dnase I digests. See page 5902.

Provisional Double Patenting

27. The nonstatutory double patenting rejection is based on a judicially created doctrine grounded in public policy (a policy reflected in the statute) so as to prevent the unjustified or improper timewise extension of the "right to exclude" granted by a patent and to prevent possible harassment by multiple assignees. See *In re Goodman*, 11 F.3d 1046, 29 USPQ2d 2010 (Fed. Cir. 1993); *In re Longi*, 759 F.2d 887, 225 USPQ 645 (Fed. Cir. 1985); *In re Van Ornum*, 686 F.2d 937, 214 USPQ 761 (CCPA 1982); *In re Vogel*, 422 F.2d 438, 164 USPQ 619 (CCPA 1970);and, *In re Thorington,* 418 F.2d 528, 163 USPQ 644 (CCPA 1969).

A timely filed terminal disclaimer in compliance with 37 CFR 1.321(c) may be used to overcome an actual or provisional rejection based on a nonstatutory double patenting ground provided the conflicting application or patent is shown to be commonly owned with this application. See 37 CFR 1.130(b).

Effective January 1, 1994, a registered attorney or agent of record may sign a terminal disclaimer. A terminal disclaimer signed by the assignee must fully comply with 37 CFR 3.73(b).

28. Claims 1-4, and 10-15 are provisionally rejected under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 123-142, and 147-152 of US copending Application No. 09/844,501 (US App. Pub. No. 20020081603).

An obviousness-type double patenting rejection is appropriate where the conflicting claims are not identical, but an examined application claim is not patentably distinct from the reference claims because the examined claim is either anticipated by, or would have been obvious over, the reference claims. See, e.g., *In re Berg*, 140 F.3d 1428, 46 USPQ2d 1226 (Fed. Cir. 1998); *In re Goodman*, 11 F.3d 1046, 29 USPQ2d 2010 (Fed. Cir. 1993); *In re Longi*, 759 F.2d 887, 225 USPQ 645 (Fed. Cir. 1985). Although the conflicting claims are not identical, they are not patentably distinct from each other.

This is a provisional obviousness-type double patenting rejection because the conflicting claims have not in fact been patented.

Claims 1-4, and 10-15 of the instant application are drawn to a method of isolating a collection of polynucleotides corresponding to the accessible regions of cellular chromatin involving treating cellular chromatin with a probe that reacted with the accessible region of chromatin, fragmenting the treated chromatin to produce marked and unmarked polynucleotides, isolating and/or cloning the marked polynucleotide fragments which correspond to the accessible region.

Claims 123-142, and 147-152 of US copending Application No. 09/844,501 are drawn to a method of preparing a library of regulatory DNA sequences. The method comprises contacting

nuclei/cellular chromatin with a first enzyme, where the first enzyme reacts with the chromatin, contacting the treated chromatin with a second enzyme to generate a plurality of DNA fragments, and cloning the DNA fragments.

Clearly, "contacting" cellular chromatin with a first "enzyme" as a probe is a species of "treating" cellular chromatin with a generic "probe" in claims 1-4, and 10-15 of the instant application. And cloning the fragments actually is actually isolating the fragments. Thus, claims 1-4, and 10-15 are anticipated by claims 123-142, and 147-152 of US copending Application No. 09/844,501.

The U.S. Patent and Trademark Office normally will not institute an interference between applications or a patent and an application of common ownership (see MPEP § 2302). Commonly assigned US application 09/844,501, discussed above, would form the basis for a rejection of the noted claims under 35 U.S.C. 103(a) if the commonly assigned case qualifies as prior art under 35 U.S.C. 102(e), (f) or (g) and the conflicting inventions were not commonly owned at the time the invention in this application was made. In order for the examiner to resolve this issue, the assignee can, under 35 U.S.C. 103(c) and 37 CFR 1.78(c), either show that the conflicting inventions were commonly owned at the time the invention in this application was made, or name the prior inventor of the conflicting subject matter.

A showing that the inventions were commonly owned at the time the invention in this application was made will preclude a rejection under 35 U.S.C. 103(a) based upon the commonly assigned case as a reference under 35 U.S.C. 102(f) or (g), or 35 U.S.C. 102(e) for applications filed on or after November 29, 1999.

29. Claims 5-9, and 16-26 are provisionally rejected under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 123-124, 128-130, 134-143, 145-147, and 151-152 of copending Application No. 09/844,501.

An obviousness-type double patenting rejection is appropriate where the conflicting claims are not identical, but an examined application claim is not patentably distinct from the reference claims because the examined claim is either anticipated by, or would have been obvious over, the reference claims. See, e.g., *In re Berg*, 140 F.3d 1428, 46 USPQ2d 1226 (Fed. Cir. 1998); *In re Goodman*, 11 F.3d 1046, 29 USPQ2d 2010 (Fed. Cir. 1993); *In re Longi*, 759 F.2d 887, 225 USPQ 645 (Fed. Cir. 1985). Although the conflicting claims are not identical, they are not patentably distinct from each other.

Claims 5-9, and 16-26 of the instant application and claims 123-124, 128-130, 134-143, 145-147, and 151-152 of copending Application No. 09/844,501 are both directed to a method of isolating a collection of polynucleotides corresponding to the accessible regions of cellular chromatin involving treating cellular chromatin with an enzyme as a probe that reacts with the chromatin, fragmenting the treated chromatin, and isolating and/or cloning the polynucleotide fragments which correspond to the accessible region. The claims differ in that claims 5-9, and 16-26 of the instant application requires that the enzyme be a methylase whereas claims 123-124, 128-130, 134-143, 145-147, and 151-152 of copending Application No. 09/844,501 recite a generic enzyme (the first enzyme) as a probe. The portion of the specification in 09/844,501 that supports the recited enzyme includes embodiments that would anticipate claims 5-9, and 16-26 of the instant application. The specification in 09/844,501 specifically discloses that a variety of enzymes can be used as probes for modifying the accessible regions of chromatin which is

known to comprises regulatory sequences, and DNA methylase is an example of such enzymes. See page 30, lines 13-34.

Claims 5-9, and 16-26 of the instant application cannot be considered patentably distinct over claims 123-124, 128-130, 134-143, 145-147, and 151-152 of copending Application No. 09/844,501 when there is a specifically disclosed embodiment in 09/844,501 that supports claims 123-124, 128-130, 134-143, 145-147, and 151-152 of that patent application and falls within the scope of claims 5-9, and 16-26 herein because it would have been obvious to one having ordinary skill in the art to modify the method of claims 123-124, 128-130, 134-143, 145-147, and 151-152 by selecting a specifically disclosed embodiment that supports the claims, i.e. the methylase embodiment disclosed in 09/844,501. One having ordinary skill in the art would have been motivated to do this because that embodiment is disclosed as being a preferred embodiment within claims 123-124, 128-130, 134-143, 145-147, and 151-152.

The U.S. Patent and Trademark Office normally will not institute an interference between applications or a patent and an application of common ownership (see MPEP § 2302). Commonly assigned US application 09/844,501, discussed above, would form the basis for a rejection of the noted claims under 35 U.S.C. 103(a) if the commonly assigned case qualifies as prior art under 35 U.S.C. 102(e), (f) or (g) and the conflicting inventions were not commonly owned at the time the invention in this application was made. In order for the examiner to resolve this issue, the assignee can, under 35 U.S.C. 103(c) and 37 CFR 1.78(c), either show that the conflicting inventions were commonly owned at the time the invention in this application was made, or name the prior inventor of the conflicting subject matter.

A showing that the inventions were commonly owned at the time the invention in this application was made will preclude a rejection under 35 U.S.C. 103(a) based upon the commonly assigned case as a reference under 35 U.S.C. 102(f) or (g), or 35 U.S.C. 102(e) for applications filed on or after November 29, 1999.

Conclusion

30. No claim is allowed.

31. Any inquiry concerning this communication or earlier communications from the examiner should be directed to Shubo (Joe) Zhou, whose telephone number is 571-272-0724. The examiner can normally be reached Monday-Friday from 8 A.M. to 4 P.M. If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Michael Woodward, Ph.D., can be reached on 571-272-0722. The fax phone number for the organization where this application or proceeding is assigned is 703-872-9306. Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to Patent Analyst Tina Plunkett whose phone number is (571) 272-0549.

32. Patent applicants with problems or questions regarding electronic images that can be viewed in the Patent Application Information Retrieval system (PAIR) can now contact the USPTO's Patent Electronic Business Center (Patent EBC) for assistance. Representatives are available to answer your questions daily from 6 am to midnight (EST). The toll free number is (866) 217-9197. When calling please have your application serial or patent number, the type of document you are having an image problem with, the number of pages and the specific nature of the problem. The Patent Electronic Business Center will notify applicants of the resolution of the problem within 5-7 business days. Applicants can also check PAIR to confirm that the problem has been corrected. The USPTO's Patent Electronic Business Center is a complete

service center supporting all patent business on the Internet. The USPTO's PAIR system provides Internet-based access to patent application status and history information. It also enables applicants to view the scanned images of their own application file folder(s) as well as general patent information available to the public. For all other customer support, please call the USPTO Call Center (UCC) at 800-786-9199.

Shubo (Joe) Zhou, Ph.D.

Patent Examiner